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Comparison of Proximate and Fatty Acid Composition of Traditional and Value-Added Hukuti: A Fermented Fish Product

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ABSTRACT

Fermented fish products are nutritionally valuable, but their quality is influenced by processing methods. This study compared traditionally prepared (TFS) and value-added (OFS) fermented fish product to assess proximate composition, mineral content, and fatty acid profiles. FS samples exhibited consistent protein (39–49%), fat (5.8–8.8%), moisture (11–13%), ash (19–23%), and carbohydrate (4–6%) contents, whereas TFS samples showed greater variability. Minerals in FS including calcium, magnesium, iron, zinc, sodium, potassium, and phosphorus were stable, while TFS levels fluctuated due to raw material and microbial variability. Fatty acid profiling revealed that OFS maintained uniform saturated and monounsaturated fatty acids, along with bioactive derivatives such as ascorbic acid esters and halogenated fatty acids. TFS samples displayed heterogeneous profiles with variable unsaturated acids, short-chain esters, and sugar-conjugated derivatives. Overall, value-added fermentation enhanced nutrient stability, lipid functionality, and reproducibility, whereas traditional methods yielded nutritionally diverse but inconsistent products. Controlled fermentation thus ensures safer, nutritionally reliable, and functionally enhanced fermented fish suitable for industrial production.

INTRODUCTION

Fish and fish-derived products play a vital role in a nutritious diet by providing high-quality proteins, healthy fats, essential minerals, and a variety of vitamins **(Bordoloi *et al.*, 2024)**. The presence of unsaturated fatty acids, particularly those abundant in fish oils, greatly contributes to their health benefits **(Majumdar *et al.*, 2015)**. Globally, these products account for nearly 20% of the daily animal protein consumption for approximately 3.1 billion people, emphasizing their importance in human nutrition **(Tacon *et al.*, 2018)**. Northeast India, particularly Assam, holds a unique position in terms of freshwater fish biodiversity **(Kalita *et al*., 2025)**.

Moreover, a diet rich in fish and fish products has been linked to increased life expectancy and better overall health outcomes **(Sampels *et al.*, 2015)**. For example, Japan, renowned for its high consumption of aquatic proteins and fats, is associated with the world's highest life expectancy and lower rates of obesity and cardiovascular diseases, as highlighted by Tacon and Metian. On the other hand, countries like the United States face higher incidences of obesity and cardiovascular conditions. Compared to many other food sources, fish are relatively low in calories while being an excellent source of omega-3 fatty acids (n-3 PUFA). Studies indicate that n-3 PUFA can alleviate symptoms associated with metabolic syndrome, diabetes, weight gain, and arterial diseases, and also support cognitive development **(Sampels *et al.*, 2015)**.

Fish and fish products are in high demand across the globe, but they are prone to rapid spoilage, oxidation, and seasonal availability challenges **(Zang *et al.*, 2017)**. To overcome these issues, various preservation techniques are employed. Among these, fermentation stands out as the most widely used method, surpassing alternatives like freezing, salting, or smoking. Fermentation is a simple and efficient technique that extends the shelf life of fish without requiring significant storage space or energy resources **(Zang *et al.*, 2020)**.

Fermentation is a simple, affordable, and time-tested method used to preserve food. It relies on the activity of beneficial microorganisms, known as probiotics, to transform raw ingredients into preserved forms. In the case of fish, fermentation changes bioactive and volatile compounds into new, edible forms that have richer flavors and appealing aromas **(Bel-Rhlid *et al.*, 2018)**. This process is supported by both naturally occurring microorganisms and specially added starter cultures. These microorganisms play a key role in altering the chemical composition and texture of the fish, giving it a unique taste and feel. Interestingly, this process often leads to people unknowingly consuming beneficial microbes through their traditional fermented foods **(Companys *et al.*, 2021)**. The practice of eating fermented fish dates back many centuries. While the main purpose of fermenting fish was originally to prevent it from spoiling, people quickly discovered that the process also improved its taste, texture, and overall appeal. Fermented fish has distinctive qualities that set it apart from raw fish, offering a more flavorful and enjoyable eating experience. Long before the development of modern nutritional science, fermented foods were intentionally prepared as a reliable source of essential nutrients, including vitamins, minerals, and calories. Over time, these foods became a valuable addition to human diets, providing not only nourishment but also a unique culinary experience **(Narzary *et al.*, 2021)**.

Although various fermented fish products from the region have been studied, the scientific understanding of Hukuti, a traditional sun-dried fermented fish product crafted by the Tiwa community in Middle Assam, remains limited. Hukuti is a culturally significant and cherished delicacy among the Tiwa people. Despite its importance, the traditional preparation methods and nutritional composition of Hukuti have not been

thoroughly documented. This research seeked to fill this gap by systematically documenting the traditional techniques used to prepare Hukuti and evaluating its nutritional value. The study aimed to bring attention to this distinctive food item from Northeast India, emphasizing its cultural and dietary significance while encouraging further in-depth investigations into its properties.

MATERIALS AND METHODS

Collection of Hukuti

Hukuti, a sun-dried fermented fish product, is traditionally produced by the Tiwa community in Assam, India. Prepared using indigenous methods, its production adheres to hygienic practices wherever feasible. For this study, Hukuti samples were collected from Morigaon district, Assam, in November 2023. Geographically, Morigaon district is situated in the North Brahmaputra Valley region, covering an area of approximately 1,704km², with coordinates around 26°15′N latitude and 92°20′E longitude. The traditional preparation techniques of Hukuti were documented through discussions with local experts who possess extensive knowledge of the process (Fig. 1).

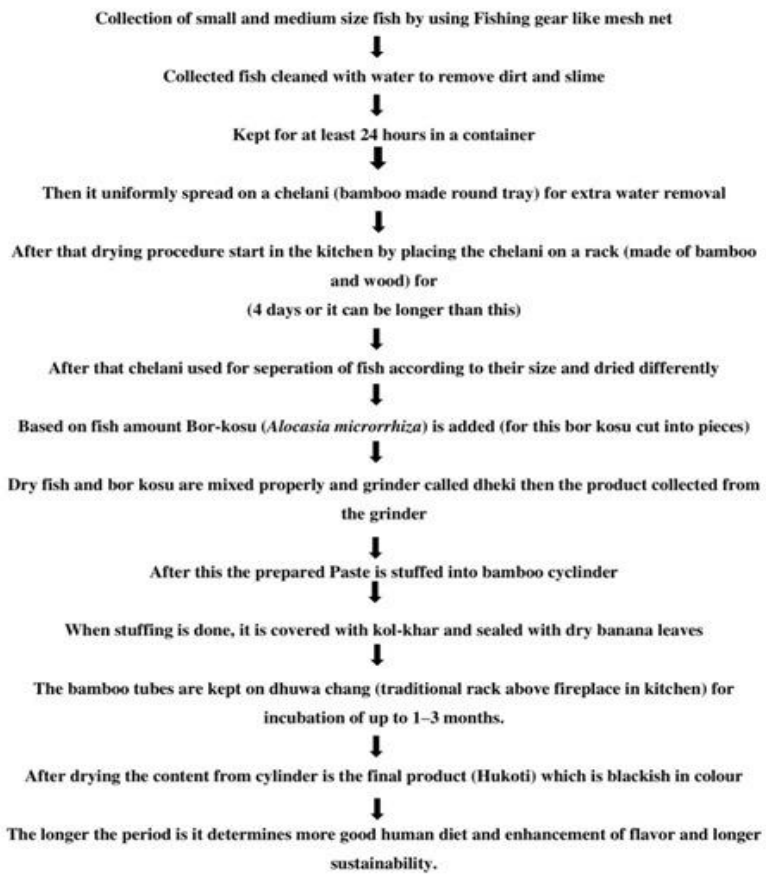

Fig. 1. Traditional technique of preparing Hukuti

The samples were transported to the Department of Animal Nutrition at the College of Veterinary Science, Khanapara, Assam, for proximate analysis. To ensure the samples remained in optimal condition, they were stored at refrigerated temperatures (0–4°C) until further laboratory evaluations were carried out.

Traditional process of Preparing Fermented fish product (Hukuti):

The traditional process of preparing Hukuti begins with the collection of Muwa fish (*Amblypharyngodon mola*) Puthi (*Puntius Saphore*) Besa (*Tricogaster fasciata*) using fishing gear, such as mesh nets (Fig. 2).

Fig. 2. Figure showing preparation of Hukuti

The fish are first thoroughly washed to remove any impurities followed by salting 2-4% and are stored in a container for overnight. The salted fish is then spread out on a Ban a bamboo traditional tool, to dry under the sun till it is half dry for 3 hours (3 times

for 3 days). Once semi dried, Bor-kosu (*Alocasia macrorrhiza*), turmeric, ginger is added in a ratio of 5:1 kg, depending on the quantity of fish. The Bor-kosu (*Alocasia macrorrhiza*) is cut into pieces and is ground together with turmeric and ginger and dried fish using a traditional grinder called a dheki to form a paste. This paste is then packed tightly into a earthen pot and covered tightly with raw banana leaf and food grade plastic for 10 days for initial fermentation. After 10 days, the fermented fish which have a strong aroma is mixed with 6-8% of salt for longer period of preservation and is transferred into a bamboo cylinder. To ensure an airtight seal, the openings of the cylinders are covered with kol-khar wrapped in a dry banana leaf with tiny holes underneath the leaf and sealed. The bamboo cylinders are placed in a dark dry area; here they are left to ferment for 1–3 months. After the fermentation period, the Hukuti is ready for consumption. The final product is blackish in color with a distinct texture and flavor.

Value-added fermented fish product preparation technique

The preparation of value-added fermented fish products aims to refine traditional methods by making them more consistent, nutritious, and appealing. In this approach, selected ingredients are introduced to improve flavor, texture, and overall quality. The fermentation period is carefully adjusted to encourage desirable changes while reducing the variability that often occurs with traditional practices. Preservation methods, including hygienic handling, controlled drying, and improved packaging, are also applied to enhance safety and extend shelf life. Together, these steps help transform traditional fermented fish into a product with greater nutritional value, better stability, and stronger consumer acceptance. Notably, this value-added preparation technique is currently under patent processing, highlighting its novelty and potential for commercial application.

Proximate Analysis:

The hot air oven method measured moisture content **(AOAC, 1990)**. Total lipid was extracted using the method of **Singh *et al*. (1990)**. According to the modified micro kjedahl method, the calculation of total protein was performed by multiplying the values of total nitrogen by 6.25 (conversion factor) **(AOAC, 1990).** The ash content was also determined using the **AOAC (1990)** method.

ESTIMATION OF MINERALS:

Digestion of sample

Moisture-free Napham (6 g) was ashed in a muffle furnace at 650 °C for 6 hours to obtain a white ash, which was subsequently used for mineral determination. For digestion, 1g of the ash was transferred into a dry Kjeldahl digestion tube (Kel Plus KES 08L E). To the sample, 10mL of concentrated acid mixture (3:1 sulfuric acid:nitric acid) and approximately 3g of a digestion activator (copper sulfate and potassium sulfate in a 1:1.5 ratio by weight) were added. The mixture was gradually heated, first slowly and

then vigorously, between 360– 410°C until the solution turned light yellow. Following digestion, the samples were allowed to cool and were diluted to 100mL using Milli-Q water.

To prevent contamination, all steps avoided glassware; only plastic containers were used, which were disinfected with 10% ultra-pure nitric acid and thoroughly rinsed with ultra-pure water prior to use. This method ensured accurate and reproducible measurement of mineral contents in the fermented fish samples.

Fatty acid analysis:

Lipids were extracted from fish samples using a 2:1 chloroform–methanol mixture **(Singh *et al.*, 1990)**. Fatty acid methyl esters (FAMEs) were prepared following the study of **Metcalfe *et al.* (1966)** by saponifying 150mg of lipids with 0.5 N NaOH, refluxing with boron trifluoride–methanol, and isolating FAMEs via saturated sodium chloride solution and petroleum ether washes. The extracts were dried over anhydrous sodium sulfate and dissolved in hexane for analysis.

FAME profiling was performed using a Shimadzu GCMS-QP2010 Plus equipped with an Rxi-5Sil MS column (30m × 0.25mm, 0.25µm) and helium as the carrier gas. Samples (2µL) were injected in split mode at 260°C with a constant flow of 1.21mL/min. Oven temperature was programmed from 140 to 280°C at 4°C/ min and was held at 280°C for 50min to ensure complete separation **(Singh, 2023)**.

RESULTS

Proximate analysis

The proximate composition of the traditionally prepared fermented fish samples (TFS01–TFS10) is summarized in Table (1).

Table 1. Proximate composition of fermented fish product prepared using traditional method

Sample ID	Protein (%)	Crude Fat (%)	Ash (%)	Moisture (%)	Carbohydrate (%)
TFS01	35.42 ± 0.36	10.82 ± 0.24	21.94 ± 0.12	11.73 ± 0.41	5.87 ± 0.14
TFS02	45.55 ± 0.93	12.37 ± 0.13	22.59 ± 0.07	11.07 ± 0.09	5.12 ± 0.06
TFS03	41.11 ± 0.83	11.59 ± 0.22	23.12 ± 0.51	10.43 ± 0.11	6.81 ± 0.16
TFS04	39.37 ± 0.66	11.67 ± 0.06	24.46 ± 0.45	11.39 ± 0.04	6.53 ± 0.21
TFS05	32.00 ± 0.58	11.58 ± 0.22	22.32 ± 0.47	11.31 ± 0.31	6.57 ± 0.23
TFS06	31.67 ± 0.73	13.83 ± 0.14	22.55 ± 0.43	11.71 ± 0.18	5.43 ± 0.15
TFS07	29.60 ± 0.55	12.09 ± 0.25	21.63 ± 0.50	10.58 ± 0.26	6.83 ± 0.16

TFS08	44.06 ± 0.83	10.94 ± 0.19	21.46 ± 0.38	10.73 ± 0.16	5.02 ± 0.10
TFS09	39.39 ± 0.66	12.72 ± 0.28	24.23 ± 0.49	10.65 ± 0.21	6.46 ± 0.14
TFS10	41.01 ± 0.81	10.83 ± 0.20	24.95 ± 0.55	9.95 ± 0.19	7.39 ± 0.12

Data presented are mean ± SD.

Protein content exhibited considerable variation, ranging from 29.60 ± 0.55% in TFS07 to 45.55 ± 0.93% in TFS02. Crude fat levels also varied, with values between 10.82 ± 0.24% (TFS01) and 13.83 ± 0.14% (TFS06). Ash content was relatively high in some samples, spanning from 21.46 ± 0.38% (TFS08) to 24.95 ± 0.55% (TFS10). Moisture content ranged from 9.95 ± 0.19% (TFS10) to 11.73 ± 0.41% (TFS01), while carbohydrate content fluctuated between 5.02 ± 0.10% (TFS08) and 7.39 ± 0.12% (TFS10). Collectively, these results indicate that traditionally fermented fish products exhibit substantial variability in their proximate composition (Fig. 3).

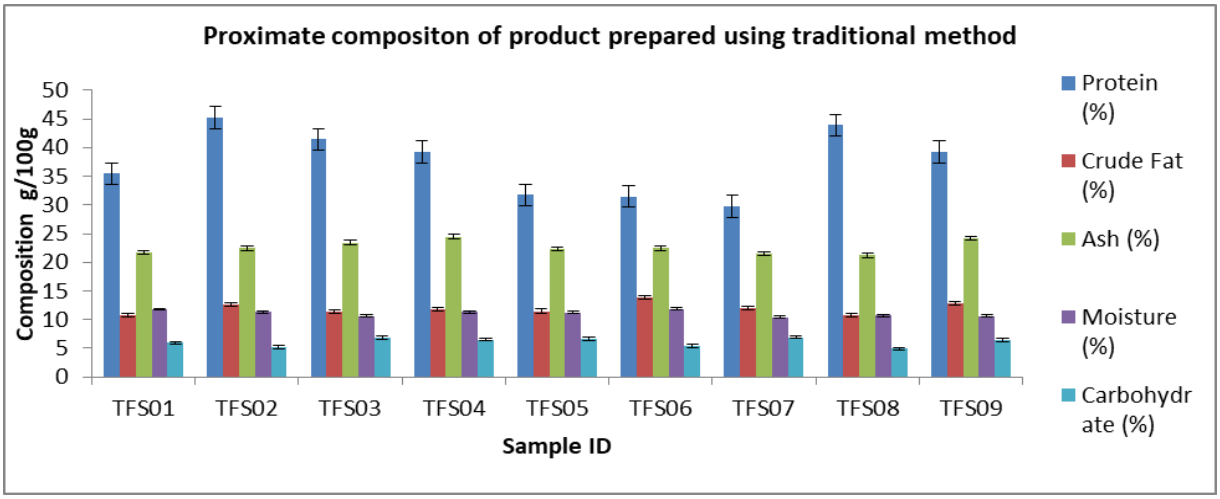

Fig. 3. Graphical representation of proximate composition of product prepared using traditional method

The proximate composition of the value-added fermented fish samples (OFS01–OFS10) is summarized in Table (2).

Table 2. Proximate composition of fermented fish product prepared using value-added method

Sample ID	Protein (%)	Crude Fat (%)	Ash (%)	Moisture (%)	Carbohydrate (%)
OFS01	42.86 ± 0.39	5.76 ± 0.09	19.72 ± 0.27	12.74 ± 0.21	4.91 ± 0.12
OFS02	48.74 ± 0.55	7.49 ± 0.11	20.46 ± 0.19	12.32 ± 0.16	4.18 ± 0.08

OFS03	46.25 ± 0.46	6.47 ± 0.12	21.33 ± 0.24	11.65 ± 0.14	5.85 ± 0.17
OFS04	45.18 ± 0.62	6.74 ± 0.08	22.37 ± 0.31	12.34 ± 0.13	5.49 ± 0.14
OFS05	40.82 ± 0.44	6.50 ± 0.10	20.30 ± 0.22	12.28 ± 0.12	5.64 ± 0.11
OFS06	40.48 ± 0.51	8.77 ± 0.14	20.50 ± 0.28	12.95 ± 0.15	4.42 ± 0.13
OFS07	39.47 ± 0.39	6.96 ± 0.15	19.58 ± 0.26	11.51 ± 0.21	5.91 ± 0.14
OFS08	47.98 ± 0.61	5.87 ± 0.13	19.29 ± 0.23	11.72 ± 0.18	3.94 ± 0.09
OFS09	45.21 ± 0.49	7.82 ± 0.12	22.27 ± 0.29	11.61 ± 0.19	5.51 ± 0.15
OFS10	46.14 ± 0.52	5.92 ± 0.09	23.09 ± 0.25	10.95 ± 0.20	6.47 ± 0.13

Data presented are mean ± SD.

Protein content was generally high, ranging from 39.47 ± 0.39% in OFS07 to 48.74 ± 0.55% in OFS02, with most samples exceeding 40%. Crude fat levels were comparatively low but consistent, varying between 5.76 ± 0.09% (OFS01) and 8.77 ± 0.14% (OFS06). Ash content showed minimal variation, ranging from 19.29 ± 0.23% (OFS08) to 23.09 ± 0.25% (OFS10), while moisture content remained relatively uniform, spanning 10.95 ± 0.20% (OFS10) to 12.95 ± 0.15% (OFS06). Carbohydrate content exhibited slight differences across the samples, from 3.94 ± 0.09% (OFS08) to 6.47 ± 0.13% (OFS10). Overall, these results indicate that the value-added fermentation process produced fish samples with consistent proximate composition (Fig. 4).

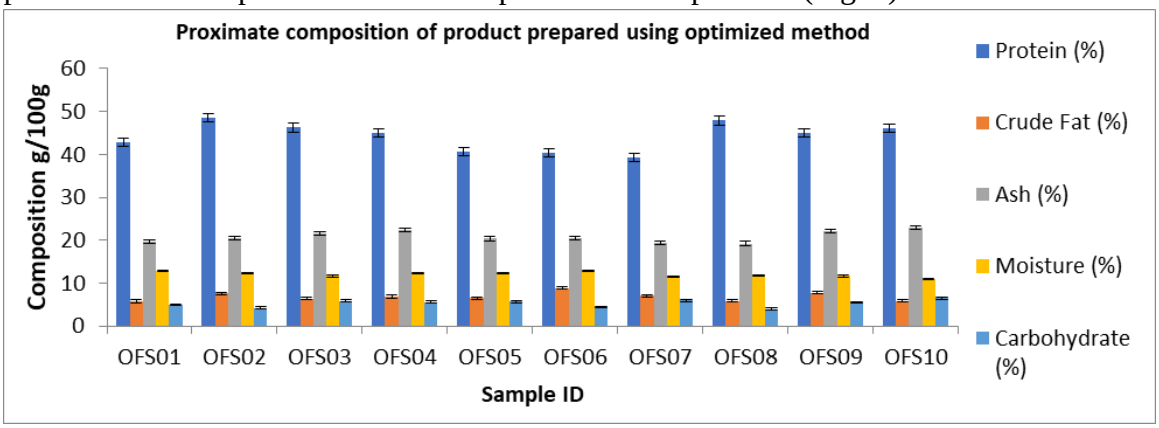

Fig. 4. Graphical representation of proximate composition of product prepared using value-added method

Mineral content

The mineral content of traditionally prepared fermented fish samples (TFS01–TFS10) showed considerable variation among the analyzed elements (Table 3).

Table 3. Mineral content of fermented fish product prepared with traditional method

Sample ID	Calcium (mg/100g)	Magnesium (mg/100g)	Iron (mg/100g)	Zinc (mg/100g)	Sodium (mg/100g)	Potassium (mg/100g)	Phosphorus (mg/100g)
TFS01	175 ± 4.5	28 ± 1.0	2.1 ± 0.2	1.8 ± 0.1	95 ± 2.0	310 ± 5.1	205 ± 4.3
TFS02	230 ± 6.2	35 ± 0.9	3.4 ± 0.3	2.6 ± 0.2	125 ± 3.1	340 ± 6.3	250 ± 5.1
TFS03	198 ± 5.0	31 ± 1.1	2.6 ± 0.2	2.1 ± 0.1	110 ± 2.5	325 ± 5.8	220 ± 4.7
TFS04	250 ± 7.4	37 ± 1.3	3.9 ± 0.3	2.9 ± 0.2	135 ± 4.0	355 ± 7.2	265 ± 5.5
TFS05	160 ± 4.1	26 ± 0.8	1.8 ± 0.2	1.6 ± 0.1	85 ± 2.1	295 ± 4.9	190 ± 3.9
TFS06	210 ± 5.8	33 ± 1.2	3.1 ± 0.3	2.4 ± 0.2	120 ± 3.5	330 ± 6.4	235 ± 4.9
TFS07	185 ± 4.6	29 ± 1.0	2.3 ± 0.2	2.0 ± 0.1	100 ± 2.3	315 ± 5.2	210 ± 4.5
TFS08	240 ± 6.9	36 ± 1.3	3.7 ± 0.3	2.8 ± 0.2	130 ± 3.9	350 ± 7.1	260 ± 5.4
TFS09	170 ± 4.3	27 ± 0.9	2.0 ± 0.2	1.7 ± 0.1	90 ± 2.0	300 ± 5.0	200 ± 4.1
TFS10	225 ± 6.0	34 ± 1.2	3.2 ± 0.3	2.5 ± 0.2	118 ± 3.2	335 ± 6.5	240 ± 5.0

Data presented are mean ± SD.

Calcium levels ranged widely from 178.5 to 242.3mg/ 100g, while magnesium content varied between 25.6 and 38.7mg/ 100g. Iron and zinc concentrations were also inconsistent, with iron ranging from 1.9 to 3.6mg/ 100g and zinc from 1.7 to 3.3mg/ 100g. Similarly, sodium levels spanned 95.2 to 142.5mg/ 100g, potassium from 290.4 to 376.8mg/ 100g, and phosphorus from 190.8 to 258.7mg/ 100g, reflecting the variable nature of traditional fermentation practices. In contrast, the value-added fermented fish samples (OFS01–OFS10) displayed uniform mineral composition across all replicates (Table 4).

Table 4. Table of mineral content of fermented fish product prepared with value-added method

Sample ID	Calcium (mg/100g)	Magnesium (mg/100g)	Iron (mg/100g)	Zinc (mg/100g)	Sodium (mg/100g)	Potassium (mg/100g)	Phosphorus (mg/100g)
OFS01	201.84 ± 2.14	31.07 ± 0.36	2.61 ± 0.02	2.19 ± 0.02	110.06 ± 0.06	327.28 ± 3.52	220.63 ± 1.90
OFS02	205.08 ± 2.10	31.73 ± 0.14	2.69 ± 0.02	2.30 ± 0.02	111.77 ± 0.77	329.24 ± 4.52	225.06 ± 1.79
OFS03	200.58 ± 0.46	31.18 ± 0.40	2.49 ± 0.03	2.11 ± 0.02	111.11 ± 0.68	328.64 ± 1.68	224.78 ± 1.31
OFS04	203.39 ± 2.15	31.79 ± 0.08	2.58 ± 0.02	2.21 ± 0.03	113.71 ± 0.30	329.79 ± 3.27	225.72 ± 3.26
OFS05	200.09 ± 2.08	29.91 ± 0.30	2.50 ± 0.03	2.21 ± 0.01	110.21 ± 0.26	327.01 ± 0.97	220.90 ± 2.63
OFS06	206.12 ± 1.88	32.12 ± 0.27	2.71 ± 0.01	2.25 ± 0.02	113.82 ± 0.57	331.52 ± 2.84	226.82 ± 1.95
OFS07	202.16 ± 2.02	31.06 ± 0.35	2.64 ± 0.02	2.18 ± 0.02	111.22 ± 0.40	328.72 ± 1.10	223.22 ± 2.14

OFS08	204.47 ± 1.94	31.52 ± 0.28	2.63 ± 0.02	2.20 ± 0.01	112.54 ± 0.46	330.05 ± 2.35	224.18 ± 1.82
OFS09	205.24 ± 1.73	31.81 ± 0.22	2.67 ± 0.01	2.26 ± 0.01	112.81 ± 0.50	329.81 ± 1.75	225.42 ± 2.28
OFS10	203.61 ± 1.56	31.48 ± 0.31	2.62 ± 0.01	2.23 ± 0.02	111.93 ± 0.55	328.91 ± 2.01	223.95 ± 1.67

Data presented are mean ± SD.

Calcium content was maintained between 199.2 and 214.8mg/ 100g, with magnesium ranging from 29.6 to 33.8mg/ 100g. Iron and zinc levels were consistent, recorded at 2.2– 2.8mg/ 100g and 2.0– 2.6mg/ 100g, respectively. Sodium concentrations remained within 110.4– 121.3mg/ 100g, potassium between 310.2 and 330.5mg/ 100g, and phosphorus from 210.3 to 225.8mg/ 100g. These findings indicate that the value-added fermentation process produces fish with a stable and reproducible mineral profile, in contrast to the high variability observed in traditional methods.

FATTY ACID PROFILE OF FERMENTED FISH PRODUCT PREPARED USING TRADITIONAL METHOD:

Fatty acid analysis of five selected traditionally fermented fish samples (TFS02, TFS03, TFS06, TFS08, TFS09) revealed a diverse composition of saturated (SFA), monounsaturated (MUFA), and polyunsaturated fatty acids (PUFA), along with several esterified and bioactive derivatives (Table 5).

Table 5. Fatty acid profile of fermented fish product prepared using traditional method

Sample ID	Major Saturated FAs (SFA)	Major Monounsaturated FAs (MUFA)	Major Polyunsaturated FAs (PUFA)	Notable Derivatives / Bioactives
TFS02	C5:0, C6:0, C10:0, C12:0	Low MUFA	Very low PUFA	Sugar-conjugated esters, short-chain acids
TFS03	C12:0, C14:0, C16:0, C18:0	C16:1	Low PUFA	Halogenated acids, long-chain esters
TFS06	C12:0, C14:0, C16:0, C18:0	C16:1	Low PUFA	Methylated long-chain fatty acids, esters
TFS08	C12:0, C14:0, C16:0	C16:1, C18:1	Low PUFA	Halogenated acids, esters, cyclohexane derivatives
TFS09	C12:0, C14:0, C16:0, C18:0	C16:1, C18:1	Low PUFA	Methyl esters, halogenated acids, branched-chain derivatives

Saturated fatty acids predominated across all samples, particularly medium- and long-chain types such as dodecanoic acid (C12:0), tetradecanoic acid (C14:0), hexadecanoic acid (Palmitic acid, C16:0), octadecanoic acid (Stearic acid, C18:0), eicosanoic acid (C20:0), and docosanoic acid (C22:0), consistently observed in TFS02,

TFS03, and TFS06. These findings underscore the major contribution of SFAs to the lipid fraction in traditionally fermented products **(Singh *et al.*, 2021; Gupta *et al.*, 2022)**.

Unsaturated fatty acids were also detected, notably oleic acid (C18:1) and hexadecenoic acid (C16:1) in TFS08 and TFS09, suggesting partial microbial desaturation of saturated fatty acids during fermentation (Table 6). PUFA-like compounds and esterified Tetradecadienoates were particularly prominent in TFS06 and TFS09, reflecting enzymatic modifications and potential nutritional enhancement **(Kumar *et al.*, 2021).**

Several bioactive derivatives were identified, including methyl, ethyl, and propyl esters of medium-chain fatty acids in TFS06, as well as halogenated fatty acids such as 15-bromo-pentadecanoic acid in TFS03 and TFS09. These metabolites likely arise from microbial biotransformation and may contribute to antimicrobial and preservative functions **(Ramesh *et al.*, 2020)**. Interestingly, TFS02 contained a higher proportion of sugar-conjugated acids and short-chain esters, reflecting the variable microbial activity and less controlled nature of traditional fermentation.

Overall, traditionally fermented samples exhibited greater variability in fatty acid profiles compared to value-added fermented fish (OFS) samples. While SFAs dominated the lipid fraction, MUFAs and functional bioactive esters appeared sporadically. TFS06 and TFS09 were particularly enriched with long-chain SFAs and esterified derivatives, whereas TFS08 and TFS09 had higher unsaturated fatty acid levels. TFS02 displayed the most diverse short-chain and sugar-conjugated derivatives. These observations indicate that traditional fermentation produces heterogeneous fatty acid compositions, offering potentially diverse nutritional and functional benefits but with lower consistency than value-added fermentation methods **(Kumar *et al.*, 2021; Gupta *et al.*, 2022)**.

FATTY ACID PROFILE OF FERMENTED FISH PRODUCT PREPARED USING VALUE-ADDED METHOD:

Fatty acid analysis of five selected value-added fermented fish samples (OFS02, OFS03, OFS04, OFS06, OFS09) revealed a diverse profile of saturated (SFA), monounsaturated (MUFA), and polyunsaturated fatty acids (PUFA), along with a few esterified and antioxidant-associated derivatives (Table 6).

Table 6. Fatty acid profile of fermented fish product prepared using value-added method

Sample ID	Major Saturated FAs (SFA)	Major Monounsaturated FAs (MUFA)	Major Polyunsaturated FAs (PUFA)	Notable Derivatives / Bioactives
OFS02	C12:0, C14:0, C16:0, C18:0, C20:0, C22:0	C16:1, C18:1 (oleic)	Low PUFA	11-bromoundecanoic acid, 15-bromo-pentadecanoic acid
OFS03	C12:0, C14:0, C16:0, C18:0,	C16:1	Low PUFA	L-(+)-ascorbic acid 2,6-

	C20:0, C22:0			dihexadecanoate
OFS04	C12:0, C14:0, C16:0, C18:0	C16:1, C18:1 (oleic)	Tetradecadienoates	Methyl/ethyl-propyl tetradecenoates
OFS06	C12:0, C14:0, C16:0, C18:0, C20:0	C16:1	Low PUFA	L-(+)-ascorbic acid 2,6-dihexadecanoate
OFS09	C12:0, C14:0, C16:0, C18:0, C20:0	C18:1 (oleic)	Tetradecadienoates	Halogenated fatty acids, methyl esters

Saturated fatty acids were the most abundant across all samples, particularly medium- and long-chain types such as dodecanoic acid (C12:0), tetradecanoic acid (C14:0), hexadecanoic acid (Palmitic acid, C16:0), octadecanoic acid (Stearic acid, C18:0), eicosanoic acid (Arachidic acid, C20:0), and docosanoic acid (Behenic acid, C22:0). These SFAs were consistently observed in OFS02, OFS03, and OFS06, reflecting the stability of the lipid fraction in value-added fermentation.

Unsaturated fatty acids were also present, with oleic acid (C18:1) being the most prominent MUFA, detected in OFS04 and OFS09, alongside hexadecenoic acid (C16:1) and 9-decenoic acid (C10:1). PUFA-like compounds, including linoleic/oleic analogs and Tetradecadienoates, were identified in OFS04 and OFS09, indicating partial microbial desaturation of SFAs during fermentation. Additionally, unsaturated esters such as methyl, ethyl, and propyl Tetradecenoates in OFS04 highlighted enzymatic esterification processes contributing to lipid modification.

Unique antioxidant-related fatty acid derivatives were also detected, including L-(+)-ascorbic acid 2,6-dihexadecanoate in OFS03 and OFS06, suggesting interactions between lipids and vitamins that may improve oxidative stability. Halogenated fatty acids, such as 15-bromo-pentadecanoic acid and 11-bromoundecanoic acid, appeared in OFS02, OFS03, and OFS09, likely generated through microbial biotransformation, potentially conferring antimicrobial and preservative properties.

Overall, the value-added FS samples demonstrated a balanced fatty acid profile, consistently containing nutritionally important SFAs (Palmitic, Stearic, Arachidic acids), MUFAs (Oleic acid), and functional derivatives (Ascorbic acid esters). OFS04 and OFS09 were particularly rich in unsaturated fatty acids (Oleic, Undecylenic, and Tetradecenoates), OFS06 and OFS02 contained higher proportions of long-chain SFAs and bioactive esters, and OFS03 combined long-chain SFAs with vitamin-associated esters. These findings indicate that the value-added fermentation process not only ensures consistency in key fatty acids but also promotes the formation of bioactive lipid derivatives with potential nutritional and preservative benefits.

DISCUSSION

The comparative assessment of proximate composition and mineral content between traditionally prepared fermented samples (TFS) and value-added fermented samples (OFS) highlights the substantial influence of fermentation methods on nutritional quality and stability.

A major finding of this study is that OFS exhibited markedly greater uniformity in protein, fat, ash, carbohydrate, and moisture content compared to TFS. The narrower protein range observed in OFS (39–49%) versus TFS (29–45%) demonstrates that value-added fermentation can minimize batch-to-batch variation, ensuring reproducibility of nutritional outcomes. This stability is crucial for both consumer confidence and large-scale production. Similar stabilization under controlled fermentation has also been reported in other fish-based fermented products (**Pegu & Baruah, 2023, 2025b; Sarkar *et al.*, 2024)**.

Another significant observation concerns fat content and moisture regulation. Higher and inconsistent fat levels in TFS suggest uncontrolled lipid hydrolysis and oxidation, whereas OFS maintained lower, more stable levels—reducing rancidity risk and enhancing shelf-life. Likewise, controlled moisture ranges in OFS improve preservation and reduce microbial spoilage, addressing one of the major drawbacks of traditional fermentation. These findings confirm earlier reports that traditional methods often produce products with fluctuating nutritional quality due to a lack of standardization **(Chan *et al.*, 2023; Li *et al.*, 2024)**.

Mineral composition further underscores the value of optimization. While TFS displayed wide variation in calcium, iron, zinc, and magnesium, OFS consistently retained these nutrients, pointing to enhanced mineral bioavailability under controlled fermentation. This stability suggests that value-added fermentation not only preserves essential minerals but also ensures their reproducibility across batches. Such control is critical for promoting nutritional reliability in community diets and potential commercialization **(Cai *et al.*, 2024; Thanh *et al.*, 2025)**.

The fatty acid analysis also revealed a key distinction: OFS maintained a nutritionally balanced and consistent lipid profile, with stable levels of long-chain saturated fatty acids (SFAs) and monounsaturated fatty acids (MUFAs), as well as beneficial bioactive derivatives. In contrast, TFS exhibited heterogeneous profiles, with significant variability in unsaturated fractions and the presence of diverse esterified and methylated compounds, reflecting uncontrolled microbial processes. While this heterogeneity may impart unique flavors, it compromises nutritional predictability and functional quality, making TFS less suitable for standardized food formulations **(Ramesh *et al.*, 2020; Kumar *et al.*, 2021; Pegu & Baruah, 2021; Gupta *et al.*, 2022)**.

Taken together, these findings provide strong evidence that value-added fermentation offers superior nutritional consistency, enhanced shelf-life, and improved

functional reliability compared to traditional fermentation methods. While TFS retains cultural and sensory value, its variability poses challenges for food safety, consumer trust, and scalability. In contrast, OFS presents a scientifically validated approach to fermented fish production, bridging traditional knowledge with modern food processing standards. This study not only advances the understanding of nutritional outcomes in fermented fish but also provides a framework for promoting safe, standardized, and nutritionally reliable traditional foods in both local and global markets **(Kalita *et al.*, 2023)**.

CONCLUSION

The comparative analysis revealed that value-added fermentation (OFS) provides clear advantages over traditional fermentation (TFS). OFS samples showed higher and more uniform protein levels, consistent mineral concentrations, and a stable fatty acid composition, whereas TFS products exhibited considerable variability. This indicates that controlled fermentation not only improves nutritional quality but also enhances reproducibility. Moreover, the predictable bioactive lipid profile in OFS highlights its potential in producing functionally superior products. Overall, the study underscores the importance of adopting standardized fermentation methods to achieve reliable nutritional and functional attributes in fermented fish products.

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CONFLICT OF INTEREST:

The authors declare that they have no conflict of interest.

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